

Molecular Relatedness of The Aboriginal Groups of Andaman and Nicobar Islands with Similar Ethnic Populations

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ABSTRACT The aboriginal tribal groups living in the Andaman and Nicobar Islands are thought to be the descendants of people who were part of the early human dispersal into the Southeast Asia. However, the origin of the tribes of Andaman, the region from where the aboriginals arrived and the route of their migration are still a matter of great speculation. To explore the origin and affinities of the Andaman Islanders, we studied the polymorphism at fifteen autosomal short tandem repeat (STR) loci, mitochondrial control region sequences and eight Y chromosomal STR loci in 194 blood samples, of which 94 samples were from the Andaman Negritos (24 Great Andamanese and 70 Jarawas) and 100 Nicobarese, a Mongoloid group of Nicobar Island and evaluated their relatedness with similar ethnic groups of India, Southeast Asia and Africa. Our results clearly demonstrate that the aboriginal populations of the Andaman Islands – the Great Andamanese and the Jarawas constitute an independent cluster, separating out from all other populations selected in the study. The Nicobarese show a close affinity with the Mongoloid population of Southeast Asia. The distinct genetic identity of the aboriginal populations of the Andaman Islands and other Asian and African populations deciphered by nuclear and mitochondrial DNA diversity suggest that (i) either the aboriginals of Andaman are one of the surviving descendents of settlers from an early migration out of Africa who remained in isolation in their habitat in Andaman Islands, or (ii) they are the descendents of one of the founder populations of modern humans.

INTRODUCTION

Andaman and Nicobar Islands, a group of about 300 islands situated in the Bay of Bengal, south of Burma and governed by India, are the homelands of six tribal populations comprising of the Sentinelese, Jarawas, Onges, the Great Andamanese of the Negrito ethnic group and the Shompens and the Nicobarese of the Mongloid ethnic stock. The aboriginal populations of the

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Andaman Islands along with the Semangs of Malaysia, the Aetas of the Philippines, and a few population groups of Papua New Guinea, who morphologically resemble the African pygmies, are the remnants of the 'Negrito' populations of Southeast Asia. The origin of these tribes of Andaman, the region and the route from where they migrated to Andamans are still a matter of great conjecture. Although the little archaeological evidence recovered so far in the Negrito settlement of the Andaman Islands suggests that the antiquity of the people of Andaman Islands does not go beyond 2,200 years (Cooper 2002), it does not imply that their antiquity may not be greater. The antiquity of their collateral groups living in the Philippines, however, is estimated to be 35,000 years (Omoto 1984). According to Bellwood (1978) the Andamanese are the early Austro-Melanesian settlers of Southeast Asia and Oceania, and not closely related to the African pygmy populations. The Andamanese and other Asian Negritos share common physical features like short stature, dark skin, and peppercorn type of head hair with scanty body hair. Women are frequently characterized by the presence of steatopygeia. The tribes of Andamans remained isolated from the outside world until the establishment of a British penal settlement in 1857. The Great Andamanese were the first to come into contact with the colonial people in the late 19th century and the settlers adversely influenced their survival, while the Onges came into close contact with outside populations almost in the mid 20th century. Until recently, the Jarawas were totally isolated and unfriendly with the outsiders. The population of the Sentinel Island still remains isolated from any outside contact. The Jarawas and Sentinelese can be considered as one of the oldest existing real hunter-gatherer populations of the world.

The overall population of the Andamanese was estimated to be around 10,000 in 1779 (Dutta

1974), which sharply declined to less than half by 1858 and at present has further reduced to 400 odd individuals. Thus, they have dwindled to a size where their survival is threatened, which may cause the irretrievable loss of the wealth of biological information. Since October 1998, the increased contact between the Jarawas and the settlers, outside visitors, as well as the deforestation caused by various developmental activities over the last decade, has affected the life and culture of the Jarawas. The emergence of such new biological and cultural environments may alter the biological fitness as well as the genetic constitution of this endogamous population in future.

Anthropological studies on aboriginal populations are scanty due to their relative isolation and unfriendliness with outsiders. Anthropological and genetic studies based on classical markers suggest a close affinity between the Andaman tribes and the Negritos of Southeast Asia rather than with the African pygmies (Radcliffe-Brown 1922; Lehmann and Ikin 1954; Nei and Roychoudhury 1993; Cavalli-Sforza et al. 1994; Sarkar 1996; Thangaraj et al. 2003). Recent studies have shown that the Jarawas and the Onges have certain unique physiological and genetic features such as low profile of blood pressure, body temperature, pulse rate and very low frequency to absence of *B* gene in ABO blood group, and that the Onges have a high incidence of HbsAg (Kumar 1987; Sarkar and Sahani 2002). Biologically, linguistically, culturally and genetically the Andaman Islanders are different from other neighbouring Asian communities.

The Nicobarese, are a large Mongoloid tribal ethnic group occupying 12 islands of the Nicobar group of islands, and the Shompens are a small Mongoloid tribal population whose distribution is restricted to the last island of Nicobar. Both these Mongoloid groups are historically known to the world due to their strategic geographical position along the ancient trade route. The Nicobarese are one of the most viable populations in the Bay Island. In accordance with geographic proximity, they speak five different dialects related to the Mon-Khmer branch of Austro-Asiatic language family. They are thought to be related to the Malays, Burmese and the Thai. Identity of their ancestry was traced to the neighbouring

island populations of Southeast Asia (Mathur 1967; Sarkar 1952). Earlier studies on mtDNA have also suggested that they are genetically similar to Southeast Asian populations (Prasad et al. 2001). Their subsistence depends upon horticulture and majority of them have converted to Christianity.

The present paper explores the molecular variation at fifteen biparental STR loci, mitochondrial DNA (mtDNA) HV I and HV II regions and Y chromosomal STR markers in three predominant aboriginal populations of Andaman and Nicobar Islands and their relatedness if any, with similar ethnic populations of Southeast Asia, Africa and mainland India. This is perhaps the first study on the diversity of bi-parental STR markers in a large representative population of the Jarawas, Great Andamanese and the Nicobarese and their comparison with data reported on similar ethnic populations of Asia and Africa.

MATERIALS AND METHODS

Population and Samples: The study includes two 'Negrito' populations of Andaman Islands - the Great Andamanese and the Jarawas and one Mongoloid group - the Nicobarese of Nicobar Islands. The blood samples were collected from 194 individuals comprising of 24 Great Andamanese (13 males, 11 females); 70 Jarawas (30 males, 40 females) and 100 Nicobarese (64 males, 36 females). Brief account of the studied population, their size, distribution and representative samples are provided in Table 1 and their present day distribution and the area of sampling are presented in figure 1.

DNA Isolation: DNA was extracted from blood samples following standard phenol/chloroform procedure (Sambrook et al. 1982). The extracted DNA was quantitated using Slot Blot technique (Waye et al. 1989).

Autosomal Microsatellites: Fifteen microsatellite loci: D3S1358, THO1, D21S11, D18S51, D5S818, D13S317, D7S820, D16S539, CSF1PO, vWA, D8S1179, TPOX, FGA, Penta D and Penta E distributed on 13 chromosomes were amplified using the PowerPlex®16 system (Promega Corporation, Madison, U.S.A.). The amplified products were separated on a denaturing 5% polyacrylamide gel using the ABI Prism™ 377

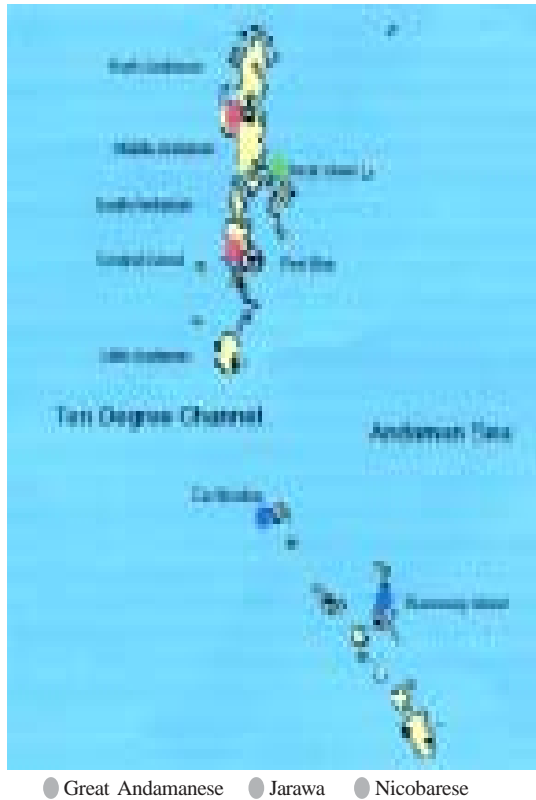


Fig. 1. Area of sampling of the aboriginals of Andaman and Nicobar Islands.

DNA Sequencer (PE Applied Biosystems, Foster City, U.S.A.). The genotypes were obtained on analysis with PowerTyper™ 16 Macro v2 (Promega Corporation, Madison, U.S.A.), GeneScan® Analysis 3.1 and Genotyper® 2.5 (PE Applied Biosystems, Foster City, CA, U.S.A.) softwares.

Mitochondrial Sequencing of HV I and HV II Regions: MtDNA sequencing of the HV I (from bases 16032 to 16378) and HV II (from bases 66 to 432) regions were carried out as reported earlier (Rajkumar and Kashyap 2003).

Y Chromosomal STR Analysis: Eight Y chromosomal microsatellite loci (DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393 and DYS385), (Kayser et al. 1997; de Knijff et al. 1997) were analysed in 107 male samples.

Statistical Analysis: Allele frequencies were computed using DNATYPE program. In addition, likelihood ratio and exact tests were performed to evaluate Hardy-Weinberg assumptions. Pairwise genetic distances (D_A) were computed using the modified Cavalli-Sforza distance measure (Nei et al. 1983) which has been found highly informative in analyzing closely related populations. Neighbour-joining phylogenies were constructed (Saitou and Nei 1987) based on the D_A measure. The first phylogenetic tree was constructed with seven microsatellites to enable comparison with African pygmies, having data on seven CODIS loci. Another phylogenetic tree was constructed based upon 15 STR loci to evaluate genetic affinities of the aboriginal populations of Andaman and Nicobar Islands with similar populations of Asia, Western Africa and mainland India. The sequences of mitochondrial HV I region of all the three studied populations were compared with suggested related ethnic populations using MEGA software.

RESULTS AND DISCUSSION

Allele polymorphisms of fifteen autosomal loci in all the three populations of Andaman and Nicobar Islands show a very narrow range in contrast to other related populations (Table 2), suggesting low diversity of the Andaman and Nicobar tribals. Interestingly, the diversity of alleles at the studied loci is observed to be much lower among the Negrito populations of Andaman Islands than in the ancient pygmy population of Central Africa (Destro-Bisol et al. 2000), which reflects that the Negrito populations of Andaman Islands have remained in isolation

Table 1: Studied populations, their distribution, population size and sample size

S.No.	Population	Ethnic Group	Area of study	Population size	Sample size	Subsistence
1	Great Andamanese	Negrito	Strait Island	44	24	Hunting gathering
2	Jarawa	Negrito	Western coast of South and Middle Andaman	250	70	Hunting gathering
3	Nicobarese	Mongoloid	Car Nicobar and Nancowry Islands	26,235	100	Horticulture

Table 2: Allele diversity in the studied STR loci among three aboriginal tribes of Andaman and Nicobar Islands and related ethnic populations.

Population	Locus															
	Parameters	D8S1338	TH01	D2S1328	D18S51	Pennt-E	D5S818	D13S317	D7S820	D16S539	CSF1PO	Pennt-D	vWA	D8S1179	TPOX	FGA
Great Andamanese	Total Alleles	5	5	7	5	10	5	6	5	7	4	7	6	6	3	7
	Frequency	68.6	27.0	27.1	47.9	22.9	29.2	41.7	66.7	20.8	39.6	41.6	60.4	52.1	72.9	27.1
Jarawa	Total Alleles	4	4	6	9	6	5	4	3	6	4	4	5	5	4	4
	Frequency	83.3	34.8	36.4	27.3	37.9	59.1	54.5	53.0	62.1	40.9	51.9	31.9	60.0	48.5	40.9
Nicobarese	Total Alleles	5	6	9	5	5	8	6	6	5	4	6	5	9	4	6
	Frequency	42.4	34.5	26.9	34.6	42.4	38.5	30.8	23.1	49.9	42.4	57.8	50.0	27.0	69.3	30.2
Bamileke	Total Alleles	8	6	13	10	NA	NA	NA	NA	NA	7	NA	9	NA	7	NA
	Frequency	30.4	50.0	28.3	19.3	NA	NA	NA	NA	NA	30.4	NA	26.1	NA	39.1	NA
Ewondo	Total Alleles	6	5	14	11	NA	NA	NA	NA	NA	9	NA	9	NA	6	NA
	Frequency	37.7	43.8	21.9	27.1	NA	NA	NA	NA	NA	29.2	NA	32.3	NA	35.4	NA
Sanga	Total Alleles	7	4	9	11	NA	NA	NA	NA	NA	7	NA	7	NA	7	NA
	Frequency	32.8	48.4	25.8	21.0	NA	NA	NA	NA	NA	21.7	NA	37.1	NA	46.9	NA
Mbenzele	Total Alleles	6	5	13	10	NA	NA	NA	NA	NA	5	NA	10	NA	5	NA
Pygmies	Frequency	30.6	52.1	23.1	22.9	NA	NA	NA	NA	NA	39.6	NA	31.6	NA	44.8	NA
West African	Total Alleles	8	6	16	17	15	8	7	7	7	7	12	11	7	7	17
	Frequency	31.0	35.0	20.0	22.0	18.0	40.5	28.5	38.0	32.0	34.5	25.0	26.0	33.5	32.5	23.0
African-American	Total Alleles	6	5	10	9	12	6	6	6	7	7	9	7	8	7	10
	Frequency	33.8	40.3	29.3	22.6	14.8	37.5	28.4	33.8	29.9	33.9	20.9	29.1	41.3	33.9	26.3
HongKong Chinese	Total Alleles	8	6	14	15	19	8	7	8	8	8	11	9	10	6	16
	Frequency	32.8	43.3	27.8	20.3	17.6	26.3	29.6	41.7	26.1	39.5	35.5	27.8	18.4	56.5	19.9
Hmar	Total Alleles	6	6	12	10	13	7	6	7	5	7	9	7	10	7	12
	Frequency	43.6	73.5	26.3	23.8	12.5	35.0	21.2	28.6	31.4	27.6	36.2	26.3	30.0	40.0	21.3
Mara	Total Alleles	7	5	10	13	15	8	6	7	7	7	10	6	9	6	11
	Frequency	41.1	62.2	18.9	16.8	13.3	28.9	26.7	35.6	22.2	34.4	32.2	32.2	27.8	40.0	16.7
Lai	Total Alleles	5	5	10	11	14	9	7	6	7	6	9	7	10	6	13
	Frequency	47.8	67.4	28.3	28.3	14.1	38.0	26.1	26.1	29.3	59.8	31.5	28.2	27.2	45.6	18.5
Lusai	Total Alleles	5	6	11	14	13	6	8	8	7	6	8	7	8	7	14
	Frequency	44.5	61.9	23.9	19.6	15.2	38.0	23.9	29.3	25.0	45.6	29.3	33.7	27.2	46.7	15.2
Juang	Total Alleles	6	6	8	10	13	6	7	7	7	6	8	6	8	5	11
	Frequency	42.0	28.0	30.0	32.0	25.0	35.0	37.0	25.0	44.0	37.0	25.0	21.0	37.0	39.0	19.0
Paroja	Total Alleles	6	6	9	11	18	6	8	6	6	5	9	7	9	5	14
	Frequency	46.8	41.0	19.2	25.0	15.1	29.5	36.5	24.4	26.0	42.9	27.3	25.0	21.1	44.2	29.3
Saora	Total Alleles	6	6	8	10	15	6	6	7	6	5	8	5	8	5	10
	Frequency	27.1	45.7	30.2	23.0	22.8	44.6	30.0	28.6	32.8	38.5	28.6	32.9	30.0	50.0	17.1

Frequency- Percentage value of the most frequent allele

NA- Data not available

for a longer period, even more than the descendents of founder populations of Africa. In addition, the observed value of heterozygosity was higher than the expected heterozygosity among the Great Andamanese and the Jarawas, suggesting these populations have experienced a bottleneck effect in the past.

All the loci in the three tribal populations were in Hardy-Weinberg equilibrium, except the FGA locus in the Great Andamanese ($p < 0.05$). This departure might be due to recent admixture of the Great Andamanese with the settlers. A rare allele 14 was observed at D16S539 locus in the aboriginal populations of Andamans, which was not found in significant frequency in any other Indian population.

The phylogenetic tree (Fig.2) constructed to evaluate the extent of genetic differentiation and the affinities of the studied populations with similar populations from Central Africa, Sanga, Mbenzele Pygmy, Bamileke and the Ewondo (Destro-Bisol et al. 2000); Southeast Asia (Law et al. 2002), two Austric speaking tribal populations, Saora, Juang and one Dravidian speaking tribe, the Paroja (Sahoo and Kashyap 2002), clearly

shows that the Jarawas and the Great Andamanese form a distinct separate cluster. This could be due to the separation of the ancestral population of these tribal groups much earlier. Although this separation is supported by a low bootstrap value (54%), we construe it to the low number of microsatellite markers employed for constructing the tree. The phylogeny constructed with 15 microsatellites for comparison with West African populations (Gonçalves et al. 2002) also suggests that the Andaman Negritos constitute a distinct cluster and separate out from the African populations (bootstrap value of 69%, Fig.3). Anthropometric, dermatoglyphic and classical serogenetic data reported earlier also suggest the similar affinities (Lehman and Ikin 1954; Sarkar 1952; Guha 1954; Sarkar 1996). Studies on the Great Andamanese based on red cell genetic abnormalities have also supported the hypothesis that they are the surviving representatives of Negrito populations that were distributed over the entire Southeast Asia in ancient times, which were genetically different from other African populations (Murhekar et al. 2001). Recent studies of mtDNA from archive

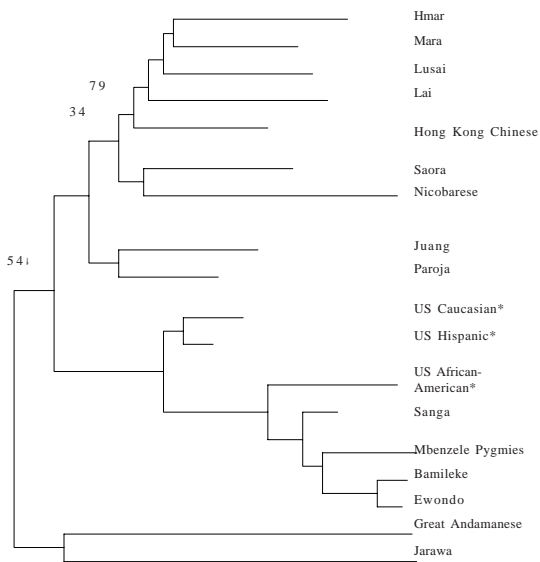


Fig. 2. Neighbour-joining phylogeny of aboriginal Andaman and Nicobar populations with other world populations constructed using D_A values based on 7 microsatellite markers.

* Levedakou et al. 2001

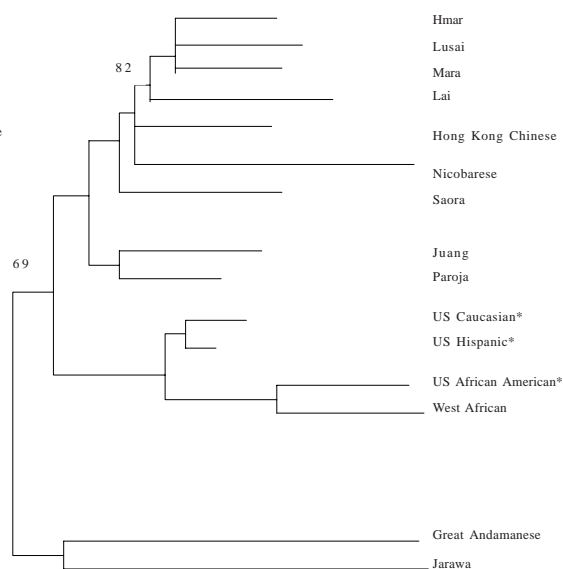


Fig. 3. Neighbour-joining phylogeny of aboriginal Andaman and Nicobar populations with other world populations constructed using D_A values based on 15 microsatellite markers.

* Levedakou et al. 2001

materials further suggest that the Andamanese are more closely related to other Asians than to modern day Africans (Endicott et al. 2003; Thangaraj et al. 2003). A southern route of migration from Africa to Australia in the Pleistocene period has also been suggested to explain the similarities among some populations in Africa, Southeast Asia and Australia (Nei and Roychoudhury 1993; Cavalli-Sforza et al. 1994; Schurr and Wallace 2002).

The neighbour-joining tree based on 15 microsatellites (Fig. 3) also reveals the Nicobarese exhibiting a close affinity with geographically proximate Indo-Mongoloid populations of Northeast India: the Hmar, Mara, Lusai and Lai (Maity et al. 2003) and the Hong Kong Chinese. The phylogenetic tree based on 7 markers further affirms the results suggesting that the Nicobarese could be the migrants of neighbouring Mongoloid populations. The Nicobarese and the Saora tribes of mainland India also exhibit a close affinity, which can be explained by their affiliation to the Austro-Asiatic linguistic family.

Thus our study clearly indicates that the Andaman tribes maintain a separate genetic identity among the world populations. The Nicobarese, however, are very much akin to other Mongoloid populations of India and Southeast Asia. Recent studies based on mtDNA variation in the Nicobarese have also suggested a close affinity with populations from mainland Southeast Asia like the Cambodians (Prasad et al. 2001).

Similar patterns of affinity have also emerged when we compared the studied populations employing mitochondrial sequences and Y chromosomal markers. In addition, a unique haplotype was observed in the mtDNA HV I region among the Jarawas with substitution at position 16086, in addition to substitutions at 16223 and 16311, already reported (Thangaraj 2003) to be predominant among the Negritos of Andaman Islands and other Asian populations. The presence of the common haplotype with substitutions at 16223 and 16311, exhibits the semblance of these populations with contemporary populations of Asia. The aboriginal populations of Andaman Islands however, seem to have remained in isolation for a much long period than any known ancient population of the world, which is evident from the presence of the

unique haplotype among the Jarawas. Mitochondrial DNA haplotype finding from archive materials of the Andamanese (Endicott et al. 2003) also substantiate the ancient origins of these populations.

In conclusion, the present study clearly demonstrates that the aboriginal populations of Andaman Islands - the Great Andamanese and the Jarawas constitute a distinct genetic pool that is different from the rest of the Asian and African populations suggesting that (i) they either are surviving descendents of early migrants from Africa who have remained isolated in their habitat in the Andaman Islands since their settlement, or (ii) they are the descendents of one of the founder populations of modern humans. The data on mtDNA and SNPs of Y chromosome are being compiled to further evaluate the origin and the antiquity as well as the route of migration if any, of the aboriginal populations of Andaman and Nicobar Islands.

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